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Porto Rico

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THE RHIZOCTONIAS OF PORTO RICO.

By J. Matz.

INTRODUCTION.

The genus Rhizoctonia is now known to be distributed in a large proportion of the gardens and fields of the world, as well as in seed beds and plant-propagating houses. Several distinct soil-inhabiting species are known as recorded in mycological and plant-disease literature. *R. violaçea* Tul. occurs more commonly in Europe. *R. Solani* Kuhn is better known in America as the sterile mycelial form of *Corticium vagum* B. & C. var. *solani* Burt. R. *microsclerotia* Matz was recently described as occuring on aerial parts of the fig tree (*Ficus carica*) in Florida, U. S. A. The last organism was found in Porto Rico during the last two years on several other hosts. From India and the Philippine Islands different distinct species have been recorded within recent years. Among the last records there are described types which are aerial; that is, they infest the above ground and even the uppermost parts of several economic plants. In Porto Rico, in addition to *R. microsclerotia,* several other forms possessing quite distinct characters both in their natural habitat and when grown in pure culture have been found, representing both groups, namely, the soil-inhabiting kinds and the aerial ones. Apparently members of the last group occur mainly in the tropical and subtropical regions. The terrestrial types or those which grow only on roots and basal parts of higher plants are perhaps as common in the warmer regions as they are in the more temperate zones, judging from the frequency with which they were encountered here in Porto Rico.

ECONOMIC CONSIDERATIONS.

The damage which the different species of the genus Rhizoctonia cause to economic plants is really large, though no exact estimate of loss is available. At first it would seem that, as compared with the more important parasitic fungi, this group might be only of secondary importance since the largest number of cases recorded are where the basal and underground parts of young seedlings were found to be attacked, thus preventing production rather than destroying crops upon which much effort has already been spent. Nevertheless the rather wide-spread occurrence of the genus in North America, Europe and Asia, and from the fact that it is prevalent in Porto Rico, and it may be justly assumed that it is present in other hitherto unrecorded regions, upon an exceedingly large variety of plants makes it one of the most important fungi affecting useful plants. Of the specific diseases attributed to Rhizoctonia there are several which are greatly instrumental in reducing the quality and amount of the crops they affect. Examples are potato Rhizoctonia blight, beet and carrot decay, bean blight, cowpea blight, fig blight and sugar-cane leaf blight and root disease. The fact that the fungus produces hardly any visible growth in the soil, and that it kills mostly the youngest roots, a casual diagnosis naturally would fail to reveal the direct agent responsible for the loss caused. Many plants are capable of regenerating new roots from near or above points of injury, in this way the root tips killed by Rhizoctonia might be replaced by new side roots as the plant keeps on growing, but even in such cases a retarding effect on the plant is inevitable, yet subsequent diagnosis may not reveal the primary cause of such a retardation.

There are certain plants which by their habit of growth and methods of cultivation become more liable to become affected by the attacks of Rhizoctonia during favorable weather conditions. Cowpeas, beans and carrots when grown in more or less wide beds or banks, producing profuse masses of foliage; sugar cane which after three or four month of growth become closed up, that is, the spaces between stools become covered by the lower and older leaves thus shading and creating a humid atmosphere around the bases of the stalks; lettuce and tomato plants when the foliage of the former and fruit of the latter come in contact with humid soil, are all liable to become infected with Rhizoctonia decay. Humid air is its chief requirement. Shade benefits the fungus only indirectly in

that it helps the highest amount of moisture to be maintained in the immediate vicinity of the tender tissues of the plant. That Rhizoctonia, or rather several members of that genus are capable of destroying the young fleshy roots of the sugar cane, a fact not recorded until recently, has been proved experimentally. Various forms of the fungus, some quite distinct from each other, were obtained, by isolation through the simple petri-dish culture method, from young cane roots on several occasions, but it is difficult to attribute directly with certainty the phenomena of stunting, "firing" of lower leaves and shortening of the joints sometimes accompanied by root development from the internodes along the stalks, to the parasitism of Rhizoctonia or any other single inconspicuous root parasite. In diagnosing such cases only the after effects of an earlier injury are observed, and the fungus which at best makes only a scanty filamentous growth has perhaps disappeared with the decomposed younger roots which were lost at an earlier stage. However, it is easy to detect those Rhizoctonias which by their nature inhabit the aerial parts of plants such as are found on the foliage and stems of the cowpea, bean, fig tree, sugar cane, and several other crops of economic value. These plants become susceptible to the latter group of fungi during periods of high humidity and at a time when they possess large amounts of tender foliage. The infected parts of the plants become soft, and finally dry, and on the decayed areas there are found, usually, numerous small, more or less rounded sclerotia. It is of interest to note that this group of Rhizoctonias though possessing in common with the less conspicuous sub-terrestrial group the distinctive mode of branching, evenness and color of mycelium as well as mode of formation, structure and color of sclerotia, yet they differ from the latter in two important respects, *i. e.*, they grow freely and produce abundant sclerotia on stems and leaves of plants, and are not causing root decay as do the members of the latter group. There is an economic significance in that. The sub-terrestrial forms are better able to perpetuate themselves by their simple filamentous mycelium under a cover of soil than those which grow above ground and are exposed to air and sun. Therefore the latter produce freely those numerous sclerotia which possess stronger powers of resistance and durability.

THE PREVALENCE OF RHIZOCTONIA IN PORTO RICO.

In a brief period of about four months various forms of the fungus were obtained in pure culture from a widely separated group

of higher plants such as sugar cane and corn, cowpeas and beans, banana, *yautía,* citrus, celery, tomato, carrot, lettuce, parsnip, beet and roselle. This does not confine the list of hosts to the above-mentioned plants alone, as no systematic search for the fungus was made. It was observed that hardly any one of the specific forms isolated were confined to a special host. It was apparent, however, that a certain preference for particular plants exists among the different kinds of the genus. On sugar cane at least three distinct species were found in widely separated localities in addition to several less markedly distinct types. Three species were found on the cowpea and bean, in addition to the common *R. solani,* at about the same time of the year in a garden, in an area of about one-eighth of an acre. Thus it is evident that in Porto Rico a larger number of Rhizoctonias are generally distributed and that they occur on a variety of host plants.

Late in 1918 it was noted that a field of cowpeas on the grounds of the Insular Experiment Station was severely attacked by a fungus which was recognized as *R. microsclerotia* and which was identical in its behavior with the fungus of the same name on the fig tree. The numerous dark brown and nearly spherical sclerotia were of the same size and structure and were produced along the petioles and tender stems of the cowpea in the same manner as they occur on the foliage and twigs of the fig tree in Florida. Pure cultures from single sclerotia were obtained for further observation and comparison. Later another organism was found possessing similar characters to the first in general behavior and relation to host but which possessed larger and fewer sclerotia. Upon culturing the two apparently distinct fungi the differences between them were even more clearly brought out in the test tube. In studying such fungi which do not produce spores readily the pure-culture method often brings out distinctive characters which are useful in determining relationships. On several occasions the original material was so fragmentary that the organisms were all-together studied from pure culture. Some of the organisms seldom, if ever, produce sclerotia on the host plant and it was necessary to resort to the pure-culture method where they could be grown in larger quantities and observe sclerotial formation. From the very beginning of the work on this group of fungi here, it was noted that the pure-culture method was essential in order to get an idea of their fixed characters. It was impossible to tell from the fragments of mycelium noted on a diseased sugar-cane root, for instance whether it is one species or

another. As a rule the generic character of even a fragment of mycelium of the genus Rhizoctonia is distinctive enough, but to determine species it was necessary to observe the sclerotia and manner of growth on a comparatively larger scale.

Ever since 1915 the writer has kept alive in pure culture two Rhizoctonias which were isolated in Florida. One is *Rhizoctonia solani* Kuhn and the other is the one which was later described as *R. microsclerotia*. These two fungi have been left at times for over six months in the test tubes which had become naturally very dry. However, the fungi were always recovered, they are alive at the present writing, and are maintaining their original individual characters. They have also been used as standards for comparison in the study of their related forms found in Porto Rico. It was found that species identical with the above two exist in Porto Rico. The above culture of *R. solani* was isolated from the underground portion of a bean seedling and compared with an authentic and identical culture of the same name sent by Dr. B. M. Duggar to Dr. C. D. Sherbakoff. The culture of *R. microsclerotia* was from type material used in its description, and which was obtained in 1915 at Gainsville, Fla., from the aerial sclerotia found on *Ficus carica*. The last named fungus was also found, in 1918, in Porto Rico on cowpeas, beans, carrots and holyhock.

An attempt to obtain a pure culture from the small sclerotia of a fungus found on cowpeas and which appeared to be similar to *R. microsclerotia,* resulted in a surprise; instead of the characteristic small sub-globose sclerotia which are invariably produced on steam-sterilized bean pods in culture tubes by the Rhizoctonia from the fig, the fungus from the cowpea produced much larger sclerotia on the same medium to the exclusion of the typical small ones. It was further observed that this difference was only found when the fungus was grown on bean pods in the culture tube, because in petri-dish cultures, using corn-meal agar, in addition to one or two larger ones the small sclerotia usually appear on the surface of the agar similar to *R. microsclerotia.* The two fungi were repeatedly and simultaneously grown from single sclerotia of about equal size and the striking difference always appeared when the fungus was transferred to the bean pods in test tubes. This difference was found to occur constantly whether mycelium or a small sclerotium was transferred to the tubes. This polymorphism of the cowpea Rhizoctonia which consists in the occurrence of small—about .5 mm. in diameter—sclerotia on the host plant and in the

petri dish, but which produces on the sterilized bean pod sclerotia of about 1 cm. in diameter, does not occur in the fig Rhizoctonia, which does not differ morphologically from the former as found in their respective host plants. A fungus identical in all respects with *R. microsclerotia,* however, was found on the cowpea and other plants on previous occasions.

Later on in the course of routinary work on plant diseases, several other more or less distinct types were encountered. Of those forms of the fungus which do not produce visible aerial growth nor sclerotia on the host plants quite a number have been isolated. Altogether there were kept for some time, in culture, forty-four different strains of Rhizoctonia. By a series of transfers of sclerotia and mycelium it was found that many of these were duplicates and only those which possessed distinct individual characteristics in form were kept. Thus it is possible that several physiologically distinct forms were discarded, but as it was not intended to make an exhaustive study of the entire subject, and to further bring out physiological points would involve extensive inoculation and cross inoculation trials for which the entire group was too large to work with, it was deemed advisable to publish an account of those which could be distinguished morphologically.

On several occasions the test-tube cultures were allowed to become dry to such an extent that when the entire contents of the tubes were placed in freshly poured corn-meal agar plates only one or two threads of mycelium developed at one point in many of the old cultures. Transfers of such single hyphæ were made by cutting off the extreme end of a mycelial thread and transferring it to either another corn-meal agar plate or sterilized bean pod in a test tube. The characters of these cultures in the newer transfers were identical with the original stock cultures in all respects and there was no indication that a tendency to extreme variation existed in the Rhizoctonias studied.

THE SCOPE OF THE WORK.

In the study of the Rhizoctonias of Porto Rico only the mycelial and sclerotial stages of the fungi were considered apart from any connection which they may have with any spore stage forms. A *Corticium* similar to *C. vagum* which is known to be the basidial stage of Rhizoctonia *soluni* was found in association with *R. microsclerotia.* (Fig. 2–A.) Cultures from the basidiospores were obtained by suspending the hymenium over an agar-poured plate. The

fallen spores were located on the surface of the agar, their germination was observed and the mycelium and sclerotia produced were identical with the typically globular and small sclerotia of *R. microsclerotia*. So far no spore stage was found in connection with the other Rhizoctonias.

Inoculations with all of the soil Rhizoctonias were made on sugar-cane roots, on the other hand the aerial members were placed on cowpea foliage and stems and it was found by reisolation that the change from aseptic to exposed conditions did not disturb the individual characters of the fungi. As already stated before, many of the fungi under consideration do not show marked specific characters as they are found on the host plants under or near the soil. By simply measuring the width of the mycelial hyphæ there could not

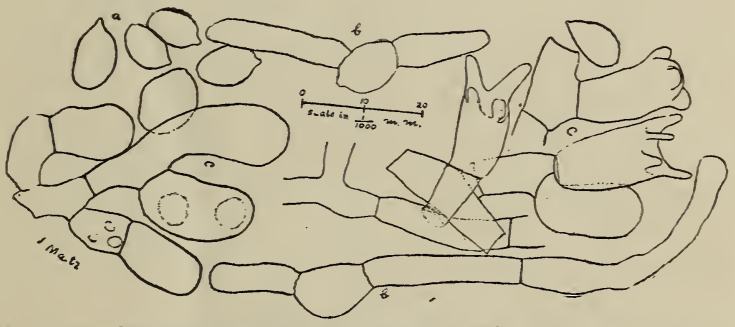

FIG. 2–A.—*Corticium* sp., basidial stage of *R. microsclerotia*. *a*, spores; *b*, germinating spores; *c*, basidia and parts of hymenium.

be obtained a fixed standard for any one in the group. Mycelium in the process of growth is much stouter and varies in thickness to a large extent. The youngest hyphæ do not furnish a basis for measurement because it was found that under certain conditions they would either tend to become thinner or stouter. To avoid possible errors it was deemed best to leave out the size of mycelium character and base classification on size and color of sclerotia as well as character of the colony growth. These were found to be constant both in the agar-plate and test-tube culture. With some of these fungi there was always noticed a change of habit in pure culture from that which they have when grown on the host plants or in the ordinary unsterilized garden soil. For instance, one of the sugar-cane Rhizoctonias produces very distinct and comparatively large sclerotia on the host plant or on the soil, but in pure culture

its sclerotia are quite insignificant. Another fungus produces rather small but numerous sclerotia on its host, one of the cowpea Rhizoctonias, yet in culture it always produces on sterilized bean or cowpea pods large sclerotia, the largest in the whole group. Such characters when they are constant throughout several generations are taken among others, as distinguishing features.

SOURCES OF MATERIAL AND METHODS OF CULTURE.

Diseased roots of sugar cane (*Saccharum officinarum*) collected from different parts of the Island furnished several strains of Rhizoctonia. Small bits of apparently diseased, reddish-brown tissue from young fleshy roots were washed in running water and placed in sterilized corn-meal agar in plates. On seven occasions the fungus was recognized by the long, stout, and straight, granular but transparent mycelium with its characteristic mode of branching and septation, to have outgrown the other fungi. With a sterilized platinum needle the extreme hyphæ from the farthest point of growth were transferred to another corn-meal agar plate. When it was ascertained that no other organism was associated with it, a further transfer was made to test tubes (three test tubes were always used for any transfer) containing sterilized green bean or cowpea pods in about 5 cc. of water. A species of *Trichoderma* and one or two other very fast-growing fungi have in some instances spoiled the chances of recovering Rhizoctonia in pure state. Another form of Rhizoctonia, quite distinct from the soil or root-inhabiting species, was grown in pure culture from the gray to brown sclerotia it produces on sugar-cane leaf sheaths and on soil in a humid atmosphere. No attempt was made to search further for Rhizoctonia on cane. As soon as it became apparent that several distinct forms of this fungus exist, instead of looking for a larger number, or to make a survey of its distribution, it was deemed best to first establish the identity of those on hand. However, in the course of the work connected with investigation on plant diseases quite a number of diseased plants have been examined and Rhizoctonia was obtained from the following plants; *beet, carrot, celery, citrus, corn, egg-plant, lettuce, pepper, celeriac, roselle, banana, field pea, tomato, Natal plum* (*Carissa grandiflora*), *cowpea, bean, yautía,* and *holyhock.* In all instances the fungus was found associated with a decay of at least a portion of the host. The method of isolation was the same in all. When the presence of the fungus was either noted or suspected in the diseased tissue of the host, bits of the diseased tissue were placed in hardened

sterilized corn-meal agar without employing the usual practice of immersing in a disinfecting liquid. As a rule when the fungus attacks a succulent root or leaf, the parasitized tissue becomes dissociated to such an extent that any liquid poison would penetrate throughout and thus kill the fungus. On the other hand the fungus is a rapid and luxuriant grower and is capable of growing away from most bacteria and fungi. In the culture plate it was noted that sclerotia formation was sometimes accelerated by the presence of some colony of the common molds or bacteria. It was also noted that the fungus does not begin to form sclerotia until the entire surface of the nutrient medium is overgrown by its mycelium, and the sclerotia are formed in places where the medium is thickest and rather farthest from the center. In many instances the sclerotia would form nearest to the rim of the petri dish. This is correlated with the fact that the terminal hyphæ are fuller with the granular protoplasm of which the older threads nearer the point of the original planting are more or less devoid. It appears that the plasma translocates itself towards the growing points and this is where the sclerotia usually originate. Apparently when growth became arrested the growing points of the plasma-filled hyphæ became distended, and instead of producing the usual long cells it produced short chains or groups of barrel-shaped cell which turned brown and became compact. These are the sclerotia. In further transferring the fungus from the petri-dish to the test-tube growth of one to two days old was used, because it was desired to carry over at least one single main branch with its shorter hyphæ rather than transferring at random. This was accomplished by holding up the plate in a vertical position against the window in the culture room, in this way in cultures one or two days old the distinct main branches stand out quite clearly. In the test tubes sclerotia are produced in a comparatively short time, with some fungi in five or six days. Here the cultures were left until they were dry to such an extent that when small portions of mycelium and sclerotia were transferred back to corn-meal agar there was no growth produced. However, the old cultures were revived by placing the entire contents of the tube in a freshly poured agar plate. This was done with the view to ascertain if age and drying out would affect a change or variation in the characters of the forms or strains under consideration. Every one of the strains were given this test at least three successive times and in this way several were lost either through dessication or by becoming contaminated too heavily

through the long stay in a dry test tube, (a minute mite would sometimes enter through the cotton plug).

While the test-tube cultures were essential for the comparison of color, form of growth and sclerotia, the petri-dish cultures served mainly to show mode of branching and manner of formation of sclerotia. Measurements of hyphæ were made from three-weeks-old petri-dish cultures.

In all the cultural work carried out here with the several members of Rhizoctonia and the frequent plantings made of single sclerotia and mycelium one interesting variation was observed. *R. microsclerotia* from bean was grown for some time in pure culture without showing any change of characters in its transfers. At one time, however, when single sclerotia of that fungus had been planted in agar it was noted that some hyphæ issuing from a sclerotium were branched at shorter intervals, and instead of producing a smooth-surfaced colony, as in the original stock culture, it produced aerial hyphæ. Transfers of this growth were made to bean pods in test tubes and the resulting growth was evidently woolly and the sclerotia were fewer and smaller. In bringing back the small sclerotia to agar plates the aerial hyphæ recurred and so a new strain has apparently sprung up. However, this phenomenon might be due to the degeneration of that particular strain, the peculiarly behaving strain does not grow at the same rapid rate as the stock culture from which it was obtained. Single sclerotia of *R. microsclerotia* from the fig strain have also produced at one time and another colonies with distinctive features. Those peculiarities consisted in a marked change in the mode of branching. (Fig. 1). In those instances the characters in the new strains were not accompanied by any evident signs of alteration in their vigor. Yet in reality it consisted of a shortening of the younger hyphæ. But the original stock culture of the fig fungus has been kept on artificial media since 1915 and it is at the present writing identical in all respects to the first isolation from which it is now many generations removed. The idea that Rhizoctonia is variable, in the sense that its characters are not fixed, can not be sustained in view of the fact that not only have the stock cultures ever lost their identity in their succeeding generation of sclerotia, but even the variants mentioned above have the inherence to maintain their peculiarities through succeeding generations. It is possible, however, that this fungus may be able to give off new forms since it usually propagates itself by budding instead of spores, *i. e.*, through its sclerotia which are only

an aggregate of buds. The density of the protoplasm in the mycelial cells may be directly influenced by the surrounding medium, thus a change of character may be maintained only as long as the new conditions are not removed. But it is not certain that new conditions could have brought about a change of behavior in the above instances. It is difficult to explain why a single sclerotium, taken from a tube culture which in turn came from a single mycelial thread, should exhibit a rather fixed change of character from its sister sclerotia planted under the same conditions and taken from the same culture tube. The physiological as well as the morphological relations between the different forms of this fungus are indeed similar. By comparing (Figs. 4, 5, 6, 7, 9) a series of fungi are noted in which a gradual instead of abrupt difference of characters exist. It would seem that a succession of changes took place at one time or another in one organism which has become divided into distinct forms which maintain their individualities.

THE GENUS RHIZOCTONIA.[1]

Rhizoctonia belongs to the group of Mycelia sterilia. *R. solani* is known to possess a basidial stage in the form of *Corticium vagum;* however, the mycelial stage of this species as well as of the others are so common and distinct that for convenience they are considered independently. There are other sterile mycelia fungi which really possess Rhizoctonia features but are known under different generic names such as *Pellicularia koleroga* on coffee trees and *Hypocnus ochraleucus* on pomaceous fruit trees. The last two fungi are now known to be the mycelial stages of two distinct *Corticiums*. When grown in pure culture they present the general features of Rhizoctonia. On their host plants they produce a brown mycelium when mature, and tend to grow in fibrils or threads. They also produce sclerotia which are similar in color and structure to those of typical Rhizoctonia. The Rhizoctonias differ from *Sclerotium Rolfsii* and *Sclerotinia* in the absence of a differenciated cortex in the former. *Sclerotium Rolfsii* differs further from the other Mycelia sterilia in that its sclerotia are smooth. There is, however, a range of variation in the form of the sclerotia and color of mycelium among the Rhizoctonias but these are not of such extremes as to place them outside the limits of the general characters of the genus.

[1] To avoid repetition a list of references to the literature dealing with Rhizoctonia is omitted here, since quite a complete list has been published by B. M. Duggar, in a paper entitled "Rhizoctonia crocorum (Pers.) D. C. and R. Solani Kuhn (Corticium vagum B & C) with notes on other Species," in Ann. Missouri Bot. Gard. 2: 408–458, 1915.

Lindau in Engler and Prantl's Pflanzenfamilien gives the characters which distinguish the genus Rhizoctonia as follows: Rhizoctonia D. C. (*Thanatophytum Nees*). Sclerotia irregular in form, often growing together, horny-fleshy with a thin not separable cortex, usually embedded in mycelium and connected by mycelial strands. Fruiting bodies are not known. Distinctions between the different species of this genus are made by other authors and are based upon the relative smoothness and size of the sclerotia, width and length of mycelial cells, and shape of cells which make up the sclerotia. In recent literature there are found references to "strains" of *R. solani,* but these mostly refer to particular sources from which the cultures were obtained, as no description, excepting one, is made of any differences in form or otherwise between the so-called strains. Rosenbaum and Shapovalof[1] described a strain of *R. solani* on potato. They found pathological, morphological and physiological distinctions in their strain.

There are really two distinct groups of Rhizoctonia, one comprises those species which inhabit and live on the aerial parts, especially foliage of higher plants, and the other contains the root-infesting species. This distinction though primarily a physiological one, also entails morphological features. The sclerotia and mycelial strands in some and sclerotia alone in others are more distinct and of a harder consistency in the aerial group. This is to be expected in fungi which are subject to the drying effect of changing air currents. There is here perhaps, evidence of evolution through adaptation to adverse conditions.

KEY TO CHARACTERS OF RHIZOCTONIAS.

Sclerotia not smooth, homogenous in structure and color, connected by mycelial fibrils ..... Rhizoctonia.
Sclerotia ærial, subglobose, brown, .2 to .5 mm. ..... R. microsclerotia.
Sclerotia ærial, subglobose, brown, .2 to .5 mm., on host plant, but larger, about 1 cm. in culture ..... R. dimorpha.
Sclerotia ærial, subglobose, .4 to 1 mm., dark brown, nearly black ..... R. macrosclerotia.
Sclerotia ærial, somewhat concave, about 2 mm., mostly gray, also brownish ..... *R.* grisea.
Sclerotia not produced on ærial portions of host plant, seldom on root crown and stem, more often in pure culture, flat, irregular, dark brown and run together to form larger coriaceous masses ..... R. solani.

[1] J. Rosenbaum and M. Shapovalof. A new strain of Rhizoctonia solani on potato. Journal of Agricultural Research, Vol. IX, No. 12, pp. 413–419, 1917; Washington, D. C.

Sclerotia not ærial, more or less regularly rounded, brown, surrounded by a lighter mycelium, often single ---------- R. ferrugena.
Sclerotia not ærial, irregular, yellowish, not coriaceous, mycellum light yellow ---------- R. palida.
Sclerotia not ærial, nearly white, irregular, mycellum hyaline --- R. alba
Sclerotia not ærial, dark brown, irregular, mycelial strands conspicuous in pure culture ---------- R. melongena.

DESCRIPTION OF SPECIES.

Rhizoctonia microsclerotia Matz. (Fig. 1–6), (Phytopathology, V. 7, No. 2, 1917.)

Vegetative hyphæ are usually .006 to .008 mm. wide with a few extremes of .004 to .011 mm. first hyaline and granular, light brown and more or less empty with maturity, septate and branched at nearly right angles.

Sclerotia superficial, both in culture and on host plant, small, .2 to .5 mm. in diameters, majority being about .3 mm., white when young, brown to dark brown at maturity, nearly homogenous in structure and color, subglobose, free from tufted mycelium, not smooth, sometimes conglomerated, abundantly produced on host plants and in culture.

Habit on cowpea, carrot, bean, *Carissa grandiflora,* and holyhock Río Piedras, P. R.

In its natural habit it is similar to several other related fungi which will be described later. A basidial stage, similar to *Corticium vagum* was found, on several occasions associated with this sterile mycelial organism, on carrot leaves and cowpea stems. Spore cultures were made and this produced a mycelium and sclerotia identical with that obtained from single sclerotia. For the purpose of identification a description of the organism based on the basidial stage would hardly be of any use as the latter occurs rarely and under special conditions.

The organism grows readily and produces a widely spreading colony in corn meal agar in petri dishes; growth is fine and transparent, at first divided in more or less distinct, plumose, main branches, later covering the entire surface of the agar culture and then first white and later turning brown, small, sclerotia are produced always on the surface of, but not imbedded in the agar. This is a feature, though not true of all the Rhizoctonias which regularly produce aerial sclerotia on the host plants, which corresponds to the habits of the fungus in its natural state.

In test-tube cultures on sterilized bean or cowpea pods growth

is fine, rapid and transparent, later becoming rarer, with numerous small sclerotia either suspended, or slightly adhering to the walls of the tube and surface of the bean pod.

This fungus was first described from Florida, U. S. A., in 1917 as occurring on *Ficus carica,* and so far there is no record of its occurrence anywhere else. In Porto Rico the organism is more common and occurs on a larger variety of plants during wet weather and it apparently lives over from one crop to another by means of its sclerotia which due to their obscure color and smallness could hardly be detected on dry vegetable matter or amongst particles of soil. It causes a decay or decomposition of the green leaf blades of cowpea, beans of several kinds, carrot and others which produce profuse foliage. On *Carissa grandiflora* it produces its sclerotia along the green but woody stems and causes the drying up of the leaves of this plant. During periods of relatively high humidity beds of cowpeas, beans and carrots were found to have been severely damage by the attacks of this fungus. The lower leaves, petioles and young stems, especially when there existed a dense top growth thus shutting off ventilation from around the bases of the plants, were in a state of slimy decomposition, and the young white and older brown, small, sclerotia were mostly found on the still green portions of the shaded parts of the plants. The way the decayed leaves adhere to the adjacent and yet unaffected leaves and petioles, by means of the fine mycelial threads, in cowpea, bean, and carrot, is characteristic of the fungus and the disease it causes. The most distinctive feature of *R. microsclerotia* is the relative small size and globose shape of its sclerotia. These remain small and true to form in pure culture and are alike there and in the field on the host plants.

As already stated before, a pure culture of *R. microsclerotia* from *Ficus carica* was kept alive in pure culture since 1915, and when compared with the similar organism isolated in Porto Rico from the several host plants mentioned previously it was found the two were identical in all respects, even to the extent of giving rise to a certain variation in the appearance of their growth. In studying the Florida fungus from the fig the following routine was carried out: A number of single sclerotia from the tube culture were transplanted back to agar plates, and when the growth was young single main branches of mycelium were transferred again to test tubes and a new crop of sclerotia were allowed to form. Some of these sclerotia were again transplanted and the process repeated so

that several complete generations were carried through. It was of interest to note that the original characters of growth, form and size of sclerotia were maintained throughout those transfers with the exception of two instances on two different occasions in cases of two colonies from two single sclerotia as shown in Fig. 1. The peculiarities noted consisted of a shortening of the side branches in one case and of a tendency to send up an aerial mycelium in the other and these were maintained in culture through several so-called generations. One of the variations noted, that of the production of aerial mycelium and resulting in a wooly appearance of the growth in test-tube cultures was paralleled in one instance in the same fungus from the cowpea isolated in Porto Rico. The sclerotia in this variant are sometimes smaller and fewer and are slower in producing mycelium when transplanted. However, it should be stated that numerous plantings of single sclerotia were made time and time again without finding any deviation from the general and typical characters of the fungus to the extent that it might be considered a variable organism. The two variations noted above may be significant in explaining why there exist today a number of distinct and yet closely related forms among Rhizoctonia.

Rhizoctonia macrosclerotia. (Fig. 7, 8 A.)

Vegetative hyphæ usually .006 to .008 mm. varying up to .014 mm. in fresh cultures, first hyaline and granular, light brown and more or less empty with maturity, septate and branched at nearly right angles, not distinguishable from *R. microsclerotia.*

Sclerotia superficial, both in culture and on host plant, .4 to 1 mm. in diameter, majority twice the size of *R. microsclerotia,* white when young, dark brown to nearly black at maturity sometimes angular, free from tufted mycelium, not smooth, not conglomerate, sparcely produced on host plant, abundant in culture.

Habit on bean (*Phaseolus* sp.) on petioles and stems, Río Piedras, Porto Rico.

Colonial growth on corn-meal agar in petri dishes is spreading and widely branching, sclerotia are produced superficially. On sterilized bean pods in test tubes the sclerotia, which are at first white and later becoming light brown to brown, are distinctly characteristic by their comparative flatness at the edges and by their larger diameter which are two times as large as *R. microsclerotia.* A short fringe of hyphal growth is usually present on the otherwise hard crust of mycelium which makes up the sclerotium. Unlike *R. miscrosclerotia,*

the inner surface of the test tube becomes plastered with the flattened sclerotia of the former which adhere to the glass more or less firmly.

The fungus was found less commonly than the previously described one. It was found first in 1919 and then again in 1921 on the same host in distinct locations, but in the same area where *R. microsclerotia* was present. The two fungi were collected from the same host though not from the same individual plant, at the same time, and were carried through in culture simultaneously. In their natural habits the two fungi are indeed very similar, they both produce sclerotia, which are similar in structure, on the surfaces of stems and petioles of a common host plant. Their differences are most pronounced when tube cultures of the two are contrasted.

Rhizoctonia dimorpha. (Fig. 8 B, 9–12.)

Vegetative hyphæ, usually .006 to .012 mm., first hyaline and granular, light brown and more or less empty at maturity, septate and branched at nearly right angles.

Sclerotia superficial, subglose both in culture and on host plant, .2 to .5 mm. diameters on host plant; in test-tube culture on sterilized bean pod, .5 to 1 cm.; two forms of sclerotia are produced on corn-meal agar in petri dishes, (*a*) small as on host plant and (*b*) larger .2 to .5 cm. in diameter.

Habit on cowpea and bean (*Phaseolus* sp.), Río Piedras, P. R.

This fungus when grown on corn-meal agar from single sclerotia or bits of mycelium produces a loosely branched and transparent colony, growth is rather thin and the main branches are plumose. Small, rounded sclerotia and one or two larger ones are usually produced in old plate cultures, they are always on the surface of the medium instead of being embedded within it. The two kinds of sclerotia are small, .2 to .5 mm. in diameter, these are smooth, spherical and dark brown, and larger spheres .2 to .5 cm. in diameter which are lighter brown and covered with a short hyphal growth. When either single small or larger sclerotia, or bits of mycelium, are transferred to sterilized bean pods in test-tube culture the resulting growth is the same regardless of origin, *i. e.*, in all the three instances large 2 to 5 mm., and sometimes reaching a diameter of 1 cm., brown, sclerotia are produced, and adhering closely to the tissue of the sterilized bean pod tissue. The development of these large masses of sclerotic fungus tissue is similar to that of the previously described Rhizoctonia; *i. e.*, they appear at first as white massess of short cells changing color to brown, they remain rather free

from overgrowth of hyphæ, are prominently raised but rather irregular in outline. When one of these large sclerotia is planted in corn-meal agar in a petri dish the mycelium after having covered the surface of the agar will produce small sclerotia, similar to those found on the host plant in nature, together with one or more of the larger types, but in the culture tubes on the sterilized bean-pod tissue not a single distinct small-type sclerotium were ever noticed among the large sclerotia massess. Single mature large sclerotia were planted in one series of agar-plate cultures and on the other hand small sclerotia were planted in a parallel series of plates using the same medium. The results in the two series of plates were identical. Bits of mycelium from the edges of the colonies in the two series were transplanted to a second double series of agar plates, and the growths here were identical in all respects. Transfers to sterilized bean pods in test tubes resulted again in the production of the larger sclerotia exclusively regardless of whether the source of the mycelium used in the transfer was a small single sclerotium or a large one.

On its host plants in the open field this fungus occurs in the form of fine mycelial strands or single threads, typical of Rhizoctonia in its manner of septation, color and branching, and it produces on the surfaces of stems, leaves and pods numerous easily detachable, small, more or less smooth, spherical, .2 to .5 mm. in diameter, light-brown sclerotia. During succeeding crops of cowpeas and beans sclerotia were picked from infected plants and planted in corn-meal agar plates and in all instances uniformly characteristic growth was produced (excepting where *R. microsclerotia* was planted, a fungus difficult to distinguish from this one in the field). At first the mycelium is transparent and grows rapidly; in about 3 to 5 days white incrustations begin to form in the test-tube cultures, soon they turn brown and increase in size, the mycelium disappears to some extent leaving the large brown incrustations, adhering to the vegetable tissue in the tube, clear to the view. The dimorphic feature of this organism, therefore, consists in its possession on the one hand in nature only small sclerotia, such as are produced by *R. microsclerotia*, and on the other hand in pure culture, unlike the last-named fungus, in addition to the small ones it possesses also distinctly larger types of uniform size, and further, on some media it produces exclusively comparatively very large sclerotia which no other of its related species possess.

The fungus was originally isolated from sclerotia and mycelium

from cowpea leaves, stems and pods, from bean leaf petioles and also from infected carrot leaves using in the last case bits of infected tissue.

The development of *R. dimorpha* on the host is more pronounced in rainy seasons and when the host plants are closely grown. The parasitism of the fungus is apparently confined to the foliage which becomes disintegrated into a mass of green slime due to the penetration of the organism between and in the leaf cell tissue. The stems, though covered with the mycelium, resist its penetration and remain green in most cases. The sclerotia which are white at first are formed on the stems, petioles and leaves. In no instance was it observed that this fungus attacks the underground portion of a plant, therefore it is not a "damping off" organism. It is capable of living over in or on the soil by means of its sclerotia and developing with remarkeble rapidity when green foliage of beans, cowpeas, or carrots are within its reach.

Inoculation tests with this organism were made with the view to prove its pathogenecity and to complete the connection between the large culturally produced sclerotia and the small ones occurring in nature. Pure cultures from bean pods in test tubes, were placed at the joints of petioles and stems of a number of cowpea plants. Shaded portions of the plants were selected, and after the inoculum was applied, they were covered with paraffined paper. In about two weeks, numerous of the typically small sclerotia were found to have developed on comparatively large areas along the stems and petioles on the locations where the inoculation material was applied. The typical destruction of the immediate leaves were noted and the fungus was reisolated from the diseased lesions and from the small brown sclerotia. From these, the large sclerotia masses have again developed in test tubes containing sterilized bean pods as well as cowpea pods. Specimens of the above diseased plants were deposited in the collection of the Insular Experiment Station at Río Piedras.

Rhizoctonia grisea (*Sclerotium griseum* Steven.) (Fig. 13, 14.)

This organism has been recorded and illustrated by W. Kruger in "Das Zuckerrohr und seine Kultur" 1899, pp. 433–466. It was also described as a Sclerotium by Stevenson in the Annual Report of the Insular Experiment Station, 1917, p. 138.

When the fungus was first grown from single sclerotia or from bits of sclerotic tissue in corn-meal agar plates it was noticed that

its mycelium possessed all the distinctive features of Rhizoctonia in mode of septation, branching, the location of the septa beyond or above the angle of branching, evenness of mycelium and transparency on the above agar and on sterilized bean pods. The formation of its sclerotia, structure and form especially when produced in pure culture are in agreement with those of Rhizoctonia in general. However it possesses such specific characters that while it agrees with Rhizoctonia more than it does with *Sclerotium* it stands out by itself sufficiently to describe it as a species of the former genus. The commonness with which the organism is encountered in Porto Rico on sugar cane, and because of its capacity to live on and destroy tender cane leaves as well as roots, warrants a study of this rather well-known fungus.

The sclerotia are gray, sometimes light brown to dark brown, not smooth, free from tufted mycelium, convex and more or less rounded dorsally, but flat and often concave ventrally, 2 to 5 m. m. in diameter, connecting fibrils very fine and hardly visible, nearly homogenous in color and structure throughout.

In pure culture the mycelium possesses, at maturity, a slight brown coloration, its sclerotia are few in number, irregular, brown yellow, flat and smaller, and are of a lesser hardy consistency than those occurring in nature. One could hardly recognize on sight a vestige of similarity between a pure culture of this organism and its form in a natural state on the host plant or on the soil. In the same measure as *R. dimorpha* seems to gather up bulk in pure culture so that its sclerotia become many times larger than they are found in nature, so does this organism lose its power for sclerotic tissue formation when it is transferred to aseptic conditions. The organism was also grown on sterilized cane leaf sheaths in test tubes, a medium on which it is mostly found in its natural habit, *i. e.* dead leaf sheaths of sugar cane, but there was hardly any sclerotia produced with the exception of a few fluffy clumps of loose mycelium. It is not known what factor might influence the production of the large, compact sclerotia of this organism in nature so that it may be artificially created when the organism grows in confinement. Like *R. solani*, it produces irregular, flat sclerotia in pure culture, but unlike the former it produces sclerotia freely in the field on the surfaces of aerial parts of its host plant. In this last point it behaves like the previously three organisms already described. It is not a *Sclerotium* because it is neither smooth, nor is it differentiated into cortex and medulla. The convex form of its sclerotia, their

form and the way they adhere to the surface of the sugar-cane leaves remind one of the mycelial form of *Hypocnus* of Pomaceous fruits (*Corticium stevensii*); however, the cane fungus does not possess the brown and conspicuous strands produced by the former. The sclerotia of the two also differ in color, those of the *Hypocnus* being uniformly dark brown at maturity. In pure culture the mycelium of the pomaceous *Hypocnus* is milky white, the colony is more compact, and it produces, in test-tube cultures rather large, though few, white wooly sclerotia. The colony growth of the cane fungus, in corn-meal agar plates, is smooth and lacks the plumose branching characters found in several other Rhizoctonias.

Reinking [1] discusses a Rhizoctonia which causes blight of the soy bean (*Glycine max* (Linn.) Merr.) in the Philippines. That Rhizoctonia possesses "roughly spherical sclerotia from 1 to 3 mm. in diameter, or they may be somewhat flattened and elongated, often 6 mm. in length." He further states that in pure culture "whitish bodies of mycelium develop which enlarge and become hard brown sclerotial bodies." These sclerotia are illustrated together with those that occur on blighted soy-bean plants and it is evident that although the author dealt with a fungus which is related in points of resemblance to one or more of the Rhizoctonias observed in Porto Rico, however, the Philippines fungus is apparently a distinct form. The size of the sclerotia and the behavior in pure culture of the latter is a combination of characters which has not been observed in the fungi thus far met in the studies of the Rhizoctonias here. For example, *R. dimorpha* produces large brown sclerotia in culture under certain conditions but on its host plant, unlike the Philippine fungus, its sclerotia are on the average less than .5 mm. in diameter and are hardly ever flattened. On the other hand the Philippine fungus possess sclerotia when on its host plants, which could fairly be compared with those of *R. grisea*, but in pure culture the sclerotia of the latter differ widely from those of the former produced under similar conditions. As stated before the cane fungus produces only small, irregular and rather soft brown-yellow sclerotic masses of fungus tissue. The mycelium and sclerotial cells illustrated by Reinking leave no doubt as to the generic community of this fungus with the Rhizoctonias of Porto Rico. Judging from the similarity, rather the identity of the basic elements of this entire group of fungi, *i. e.*, their mycelial and sclerotial structure, it would seem that their chief points of differences are really physiological or fixed behavior

[1] Otto. A. Reinking. Philippine Journal of Science, Vol. XIII, No. 4, Sec. A, 1918.

through adaptation to certain environmental conditions. Even should there be found distinct basidial forms for every one of the different Rhizoctonias, the conception that they are closely related need not be changed.

In summarizing the characters of the aerial Rhizoctonias it should be noted that none of them are associated with the damping-off of young seedling plants. There is apparently a physiological specialization in this group as contrasted with that group typified by *R. solani* which will be considered later. It seems to be a correlated fact that those Rhizoctonias which generally produce aerial sclerotia in nature do not as a rule attack the underground portions of tender plants.

There is one organism which though resembling Rhizoctonia in cell structure and habit, yet does not produce true sclerotia in culture and those which it produces in nature are so flat and small that they never take on the form of incrustations or compacted fungus tissue. The organism *Pellicularia koleroga* (*Corticium koleroga* Burt.) is here referred to. On its host plant, the coffee tree, the fungus consists of brown, smooth strands of mycelium on the trunk and twigs terminating into a white, spreading and thin web of mycelium on the underside of the green leaves which turn brown and die. This thin web of mycelium turns brown with age and may produce, sometimes quite regularly, small, flat, and brown clumps of short hyphæ which resemble a sclerotium arrested in growth at the beginning stages of its development. The fungus grows slowly in culture, and produces a transparent mycelium which turns slightly brown with age. In the culture tube the mycelial growth is rather woolly with a few clumps of loose mycelium standing out above the surface of the culture medium. There is a certain resemblance between this fungus and the *Hypocnus* of pomaceous trees, especially in cultural appearances. The formation of brown strands along the twigs of the host plants of the two fungi is another point of similarity, but they differ largerly from each other in the form of sclerotia produced in nature.

The following are descriptions of the soil-inhabiting fungi or those which are not commonly found on the aerial portions of plants. Since the characters of these fungi are brought out mostly when in pure culture, and because of the rather lack of sharp lines of distinction in some only the more clearly distinct types will be taken up. For the sake of comparison a review of the well-known soil fungus *Rhizoctonia solani* Kuhn is here given.

Rhizoctonia solani Kuhn. (Fig. 15, 16, 17.)

The septate mycelium produces a coarse, transparent growth in corn-meal agar plate cultures, growth is rapid but sclerotia are not readily produced in culture plates. In older plate cultures, however, small, irregular, flat, light-brown masses of sclerotial cells grouped in short irregular chains may be found embedded in the medium. These sclerotia occur only in a few places, especially toward the edge of the plate, and are not generally distributed. Hardly any perceptible change in color of the colony takes place on corn-meal agar, though the more mature portions of the mycelium, when seen with the microscope, are slightly brown. The younger terminal hyphæ and side branches are filled with granular protoplasm. The older and larger hyphæ become vacuolated and at length empty. It appears as if the fluid contents of the long mycelial cells gradually empties it self into the growing extremities and finally when growth is arrested either because of lack of space for expansion or because of lack of food, the fluid becomes locked up in the many terminal short cells which comprise the structure of the sclerotia. After four or five days of growth in the ordinary petri dish on corn-meal agar, the main branches become clearly vacuolated and appear to have lost their fullness. This does not indicate loss of vitality, however. When either young or old growth of mycelium is transferred to a sterilized green bean pod in a test tube, the formation of sclerotia and change of color in the mycelium from transparent to brown takes place after the entire surface of the sterilized bean-pod tissue is overgrown, by the rapidly advancing mycelium. The formation of sclerotia begins first with white, irregular clumps of short cells which soon change in color to light brown and then dark brown. At length the mycelial strands as well take on a brown coloration, they become thin and growth seems to stop. The sclerotia, as a rule, are found on the strands adhering to the walls of the glass tube as well as to the tissue of the medium. The culture may remain in this way for six months, more or less, without losing its vitality. A mature culture of this fungus possesses the following characters: sclerotia dark brown, irregular, tending to elongation, flat, and not possessing conspicuous marginal growth of mycelium. The mycelial strands are slightly coarse, but not generally straight, not conspicuous and of a lighter brown than the sclerotia.

The organism grows rapidly on moist and slightly acid vegetable media. It does not tend to vary in appearance, though its mycelium varies in width from .008 to .012 mm. Fig. 15 shows the same fungous

photographed in Florida, U. S. A., in 1915 and then again after many transfers in culture and allowing it to dry out for lapses of six months or more it was photographed in 1921. It is evident that no change has taken place in the appearance nor virulence of the organism. This statement might seem superfluous when it is taken in consideration that the organism is as old at least as any living plant, yet in view of the fact that workers on fungi often encounter what they consider variable forms, and especially when there exist several Rhizoctonias closely resembling each other it will be of interest to know that what is here described and illustrated as *R. solani* is a fungus which maintained its individual characters for a considerable time in artificial culture. The identical fungus was isolated in Porto Rico from bean pods which were partially decayed and touching the ground at Guayama, from citrus (Grapefruit seedling in pot), Roselle (*Hibiscus sabdariffa*) seedling, celery, tomato stem, lettuce leaves, sugar-cane roots, banana roots, and pea (*Pisum* sp.) at Río Piedras. In all of these the fungus was found in the mycelial stage only. This list of hosts is by no means complete, as there is no doubt that there is a larger number of plants which become infected with this fungus during certain seasons. It is of interest to note that some of the host plants mentioned above had been found to be attacked by other forms of Rhizoctonia quite distinct from each other. Thus two distinct forms were found on celery, two on lettuce, and several on the bean. These distinct forms were found in not widely separated areas. Of the Rhizoctonias found on sugar-cane roots there are several which exhibit marked distinctions from each other. Some features, though they were constant through the several transfers in pure culture yet were not deemed sufficient to determine the individuality of the organism. While one can differentiate between slightly differing cultures through continuous observation and comparative study of a given group it would only add confusion to those who may not have more than one or two kinds at one time. There were kinds of Rhizoctonia which instead of producing sharply outlined, yet irregular, sclerotia in more or less irregular strings or groups as shown in Fig. 15, produced irregular patches of sclerotia of a softer consistency and of a lighter brown color (Fig. 23). Another strain produced more or less rounded bulged sclerotia, still another produced an abundance of mycelium and only a few though distinct sclerotia. Thus in sorting out the different strains of the *R. solani* type or those which were found on roots of plants and which possess more or less flat

irregular sclerotia, only four new fungi, in addition to the true *R. solani* type, will be described in the following paragraphs.

Rhizoctonia palida. (Fig. 18, 19, 20.)

This organism is very pale yellow in color when grown under the same condition as *R. solani.* It never takes on dark brown or brown coloration in the culture tube on sterilized bean pods. Yet in its mode of septation branching and formation of sclerotia it agrees well with any of the true Rhizoctonias. In plate culture on corn-meal agar it produces a fine transparent colony, the main branches of which are plumose but which do not spread as widely as in *R. solani,* but more so than in *R. grisea.* It produces, in older plate cultures small flat and yellow sclerotia embedded in the medium. On bean-pod cultures in test tubes it makes a rather woolly growth with few small irregular and flat pale yellow sclerotia. Its mycelium is finer, measuring from .004 to .006 mm. in diameter. There is a strong resemblance between the pure cultures of this organism and *R. grisea* in their color and consistency of the sclerotia, but they differ in the following respects: in agar-plate culture the colony of the latter is straight without showing hardly any tendency to produce plumose branching; in test-tube cultures the latter produces harder sclerotia.

R. palida, in addition to its occurrence on cane roots, was also found on roots of pepper plants, *Capsicum* sp., and on young corn seedlings. This organism occurs less commonly than those which produce more resistant sclerotia. It was found in association with a decay of basal parts and young rootlets of *Capsicum* near Manatí, P. R., and with a root decay and chlorosis of *Zea mays* and sugar-cane at Río Piedras, P. R.

Rhizoctonia ferrugena. (Fig. 21, 22.)

In corn-meal agar plates, this organism produces a zonated pale white colony, rather slow growing at first, later producing sclerotia embedded in the medium, mostly nearer the outer edge of the petri dish. The sclerotia are formed from short stout hyphæ the cells of which are short and nearly barrel-shaped. This process of sclerotial formation is common to all the Rhizoctonias studied so far. In the test tube on sterilized bean-pod cultures the mycelium is transparent and maintains its transparency much longer than the other hard sclerotia-producing organism of the same group. Numerous and scattered sclerotia are produced; at first they are white; then turning yellow-reddish, later becoming rusty red, are irregular in

margin but maintain a more or less even diameter, in contrast with *R. solani* in which the sclerotia are usually elongated or grouped in only one direction. Grouping takes place in *R. ferrugena* but it is merely a closer association of individual sclerotia, in *R. solani* there is an actual conglomeration or growing together of sclerotia.

R. ferrugena was isolated from diseased sugar-cane roots in the beginning of 1919, and throughout the many transfers of this organism it maintained its distinctive features.

R. melongena. (Fig. 23, 24, 25.)

In petri-dish cultures this organism presents no distinct features by which to distinguish it from *R. solani.* It produces a rather coarse growth with dark-brown flat sclerotia in corn-meal agar. On sterilized bean pod in test tube it produces straight, long, at first hyaline, later dark brown strands at the ends of which are formed strings of irregular flat sclerotia. Usually these sclerotia accumulate towards the bottom of the tube to form dense masses or crusts of brown sclerotic fungus tissue.

The organism was isolated from decaying egg-plant (*Solanum melongena*) seedlings. These plants were collected in a garden at Río Piedras, P. R., where a large variety of other garden crops were grown and upon the roots of which true *R. solani* was found at the time.

Rhizoctonia alba. (Fig. 26, 27, 28.)

The organism produces a coarse transparent growth on corn meal agar in petri-dish culture, and it possesses a whiteness which becomes accentuated as the colony becomes older. It is the only fungus in the whole group, which though possessing a mode of septation, branching, size and general appearance of *R. solani,* but it is deficient in the yellow pigmentation which is common to a higher or lesser degree in all the Rhizoctonias under observation. The degree of yellowness possessed by the different members of this group of fungi varies from very dark brown to slightly yellow. This organism is practically without any color. It was observed that in the plate cultures where this fungus was growing a certain opaqueness in the medium took place and it became more pronounced as the colony progressed. There were numerous small crystals in the medium, apparently a direct result of the action of the organism on the substances in the corn-meal agar medium. In test-tube cultures on sterilized bean pods the growth is coarse and transparent,

later becoming white with the irregular sclerotia remaining white to slightly yellow.

R. alba was isolated from the basal parts of partly decayed leaves of *Apium* sp. The diseased plants collected at Maricao, P. R., were sickly yellow and many of their outer leaves were dead. The basal parts of some of the remaining leaves as well as the root crown itself were soft in part. The affected tissue when examined under the microscope showed that mycelium of this Rhizoctenia penetrated the interior of the affected tissues.

PUBLICATIONS OF THE YEAR (1920–21).

(PUBLISHED OR IN PRESS.)

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The Journal of the Department of Agriculture of Porto Rico, Vol. IV, No. 4. Changes Wrought in the Grapefruit in the Process of Maturation, by F. A. López Domínguez.
The Journal of the Department of Agriculture of Porto Rico, Vol. V, No. 2. The Minor Sugar-Cane Insects of Porto Rico, by G. N. Wolcott.

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Fig. 1-28

FIG. 1.—A, *R. microsclerotia* in corn-meal agar culture. B and C, variations in manner of branching sometimes found in the same fungus from the fig tree.

FIG. 2.—Bean plant attacked by *R. microsclerotia.*

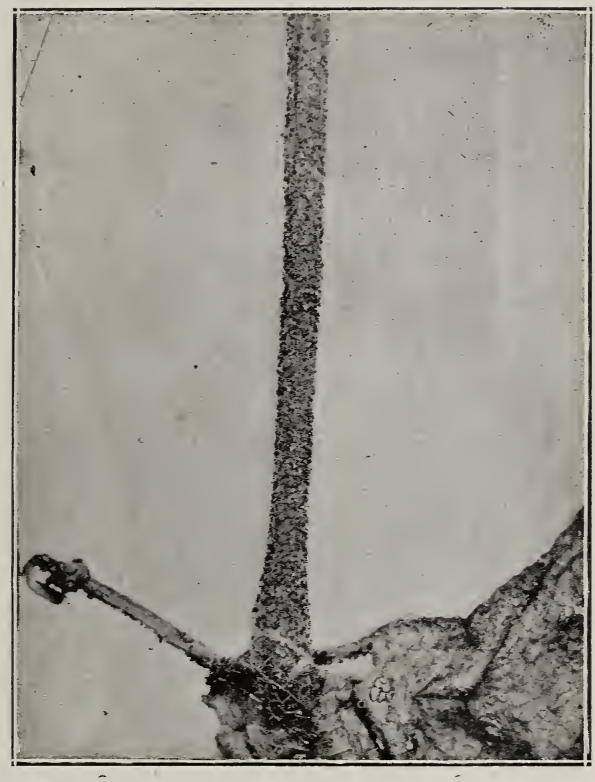

FIG. 3.—*R. microsclerotia* on cowpea. Note the destruction of foliage and the numerous minute sclerotia along the vine.

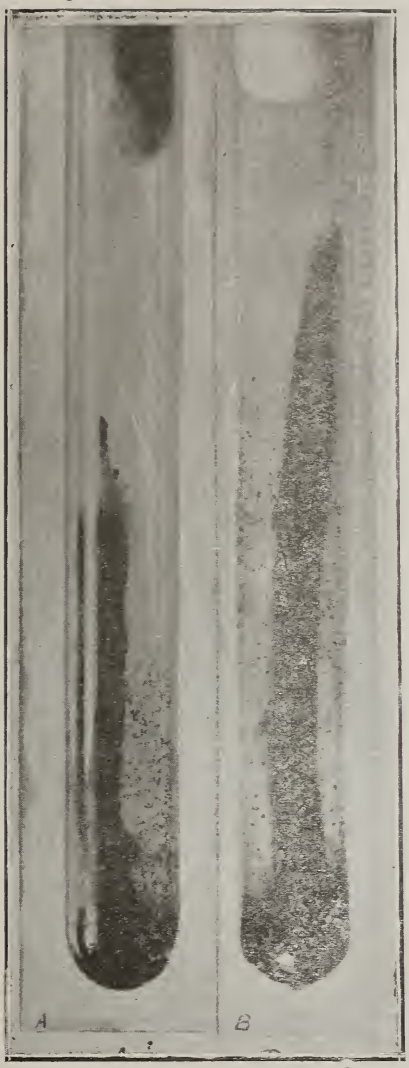

FIG. 4. — A, *R. microsclerotia* photographed in 1915. B, the same organism after six years of growth on several artificial media, photographed April, 1921. The organism was isolated from a fig tree in Florida, U. S. A., in 1915.

FIG. 5.—A, a culture of the woolly variety of *R. microsclerotia*. B, *R. microsclerotia* from cowpea, isolated in Porto Rico in 1918.

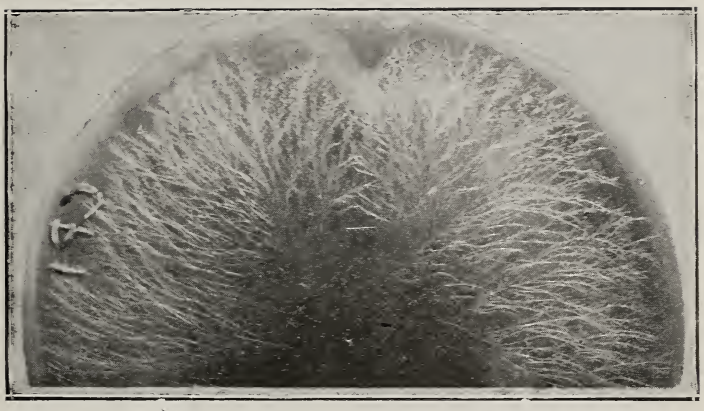

FIG. 6.—*R. microsclerotia* from cowpea. Two-days-old culture in corn-meal agar.

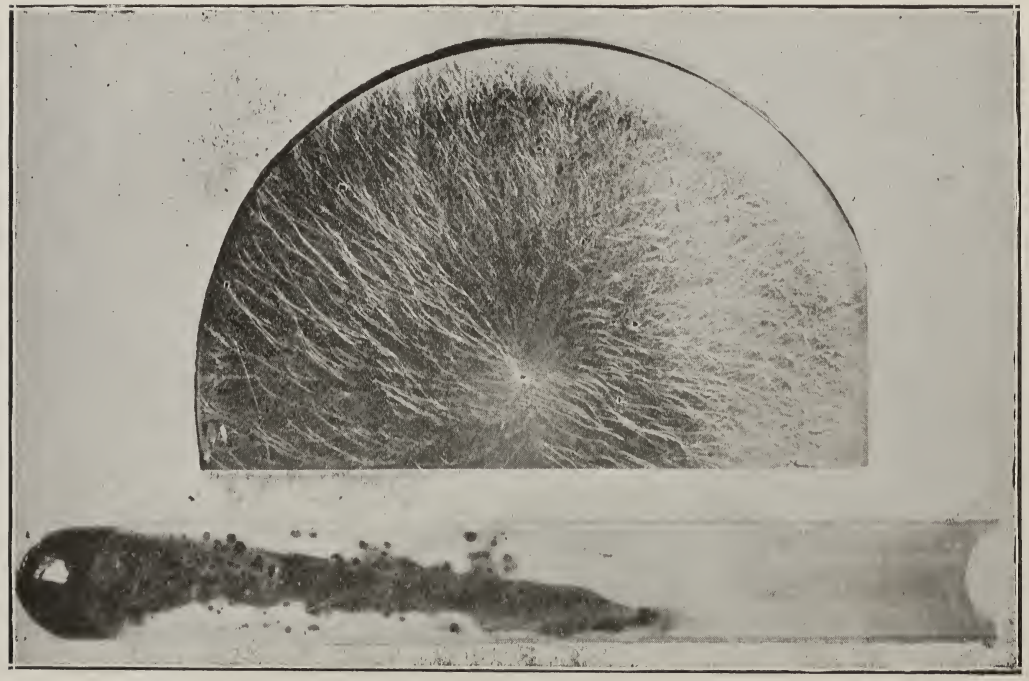

FIG. 7.—*R. macrosclerotia*, mature, culture showing growth characters on sterilized bean pod to the right. Left is a corn-meal agar culture three days old.

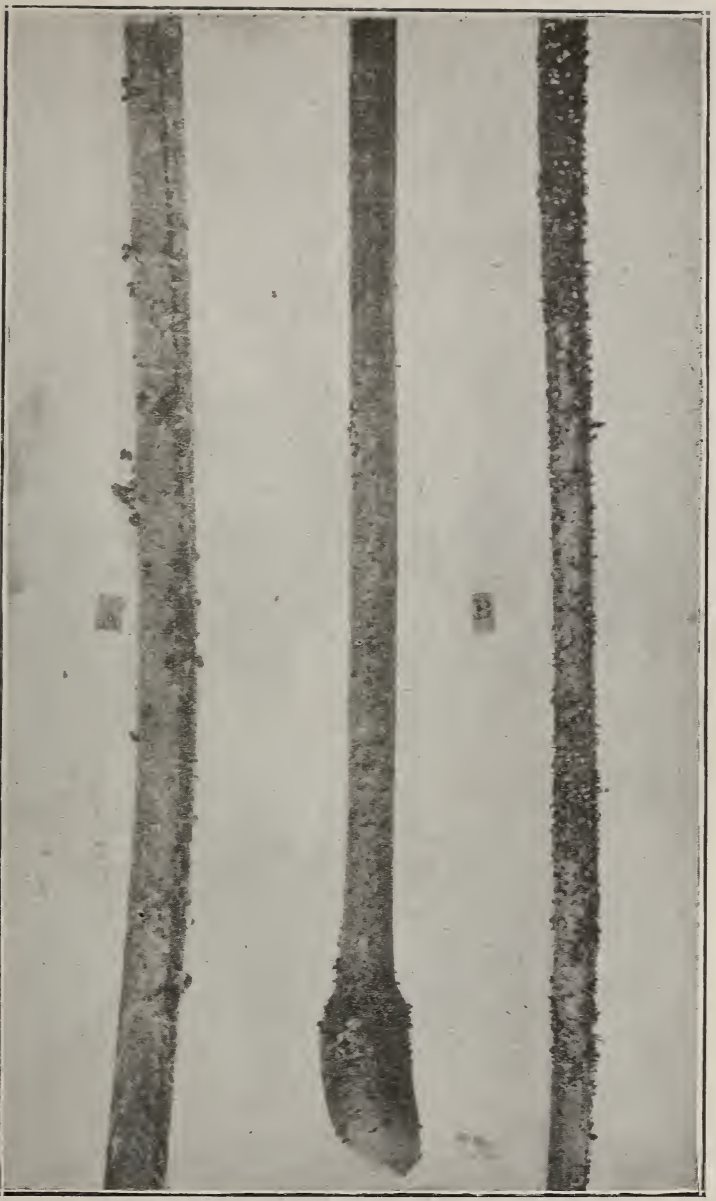

FIG. 8.—A, *R. microsclerotia* on bean vine × 2. B, *R. dimorpha* on centre and right petioles of the same host, × 2. Note the relative size of the sclerotia in A and B.

FIG. 9.—*R. dimorpha.* Tube on left is an old culture; central tube is from a large sclerotium; tube on right is from a small sclerotium. Right and center tubes are 2 weeks old.

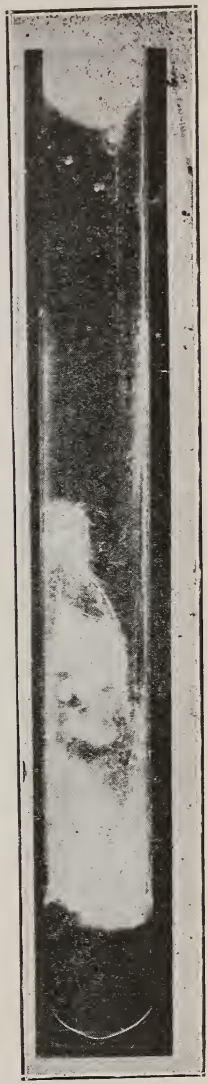

FIG. 26.—*R. alba.* Mature culture on sterilized bean pod.

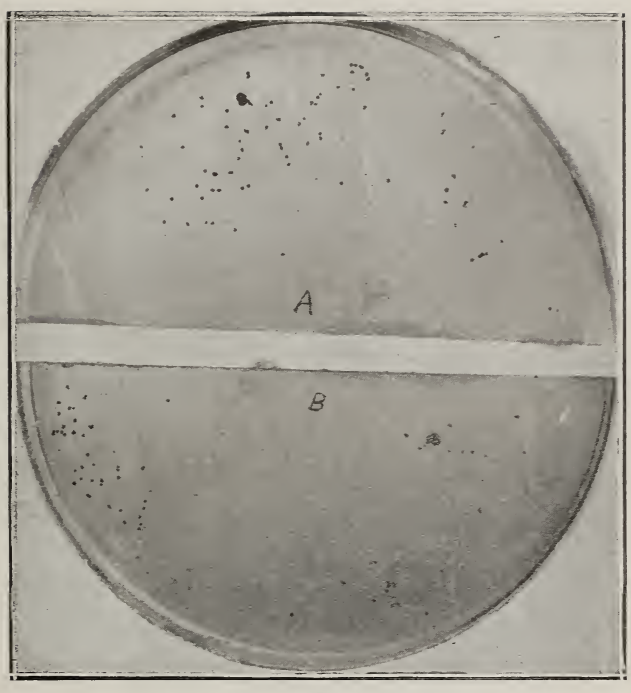

FIG. 10.—*R. dimorpha*. Old corn-meal agar cultures. A, from a small sclerotium; B, from a large sclerotium. Note the small and large sclerotia produced in both.

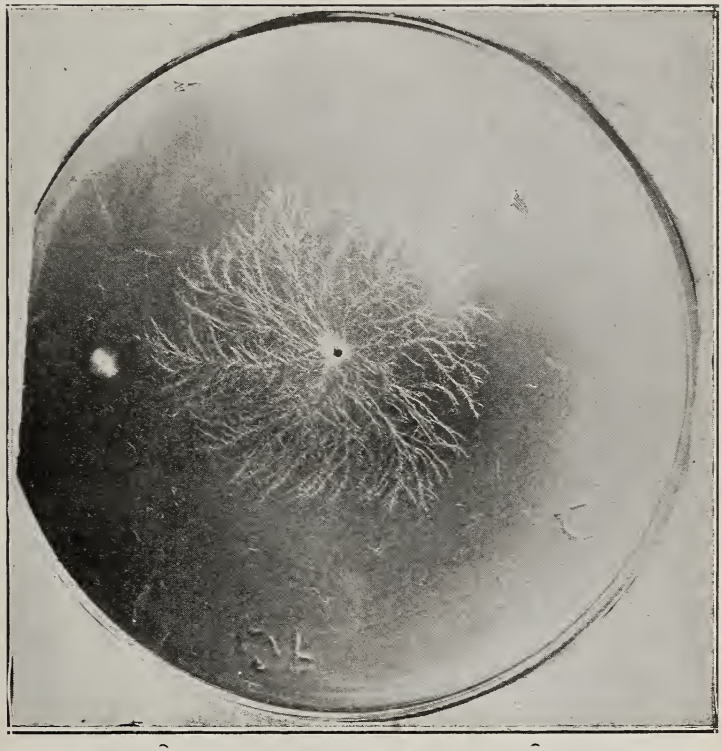

FIG. 11.—*R. dimorpha.* Two-days-old corn-meal agar culture.

FIG. 12.—*R. dimorpha* on bean.

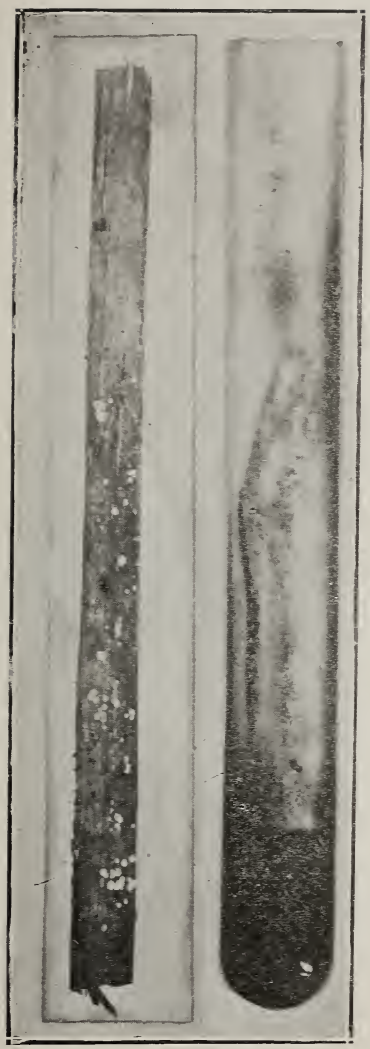

FIG. 13.—*R. grisea.* On right is a pure culture of the organism obtained from the gray sclerotia on the cane leaf to the left.

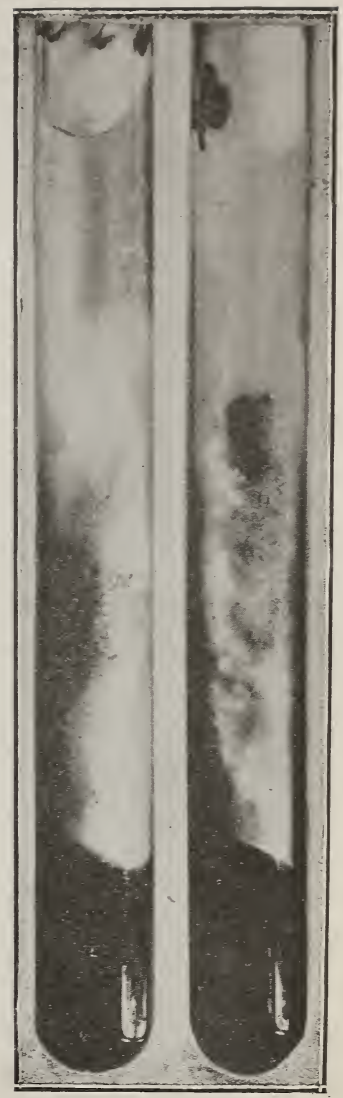

FIG. 18.—*R. palida.* Mature cultures on bean pods.

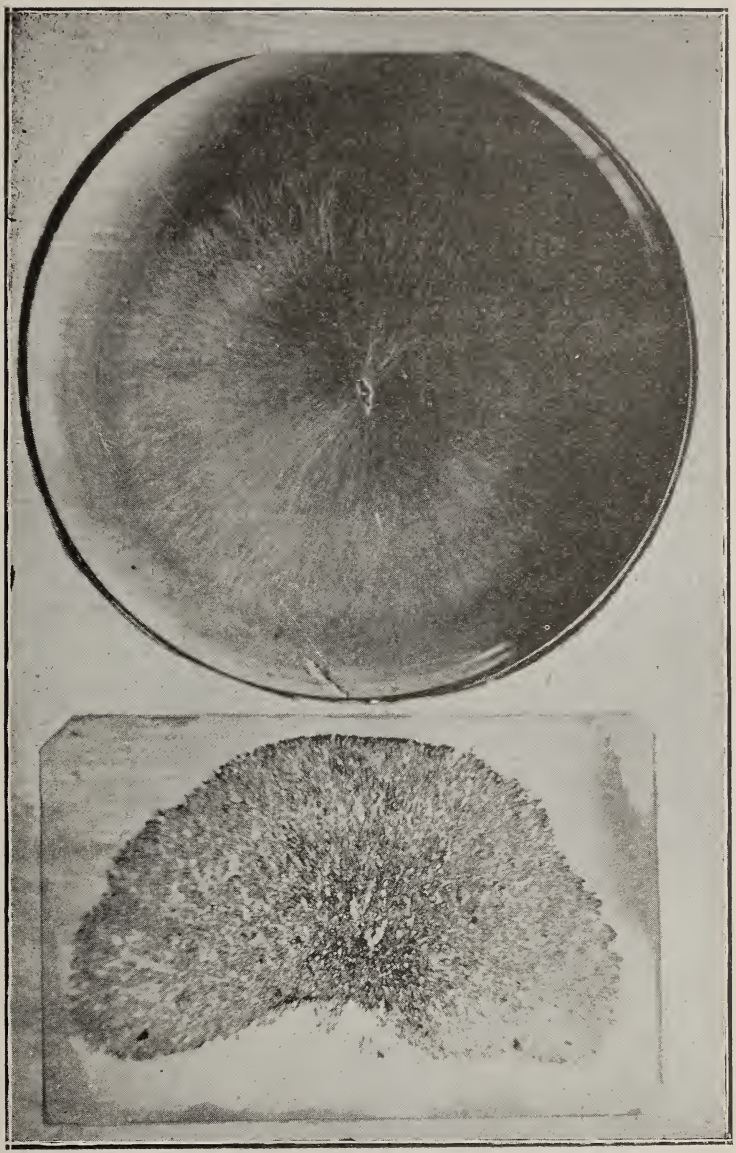

FIG. 14.—Above is a culture of *R. grisea* on corn-meal agar, three days old. Below is a cross section through a mature sclerotium of the same fungus, enlarged × 50.

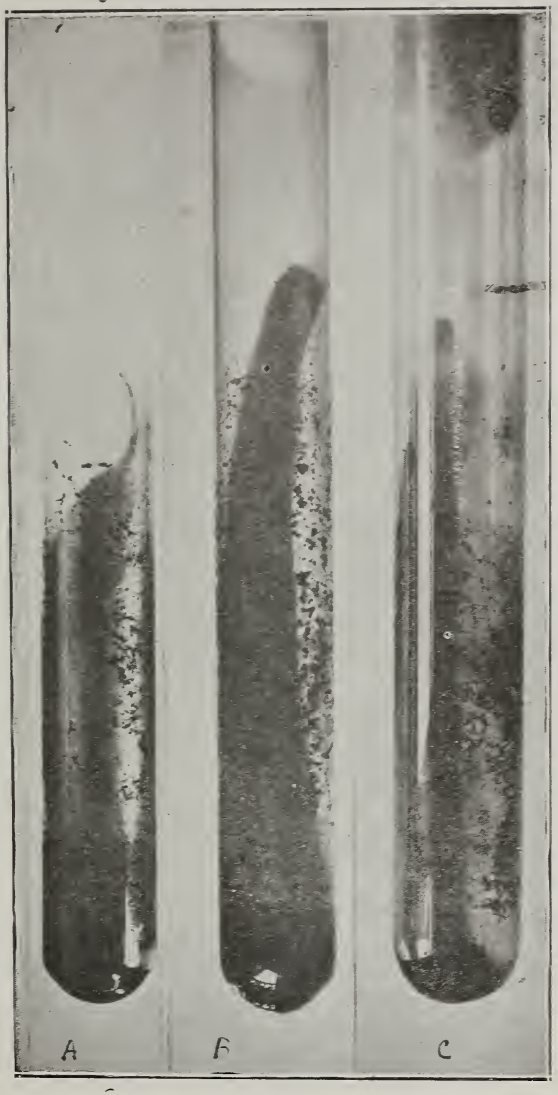

FIG. 15.—C, *R. solani* Kuhn photographed in 1915 in Florida; B, the same organism after six years of growth in several artificial media, photographed April 1920; A, *R. solani* isolated in Porto Rico.

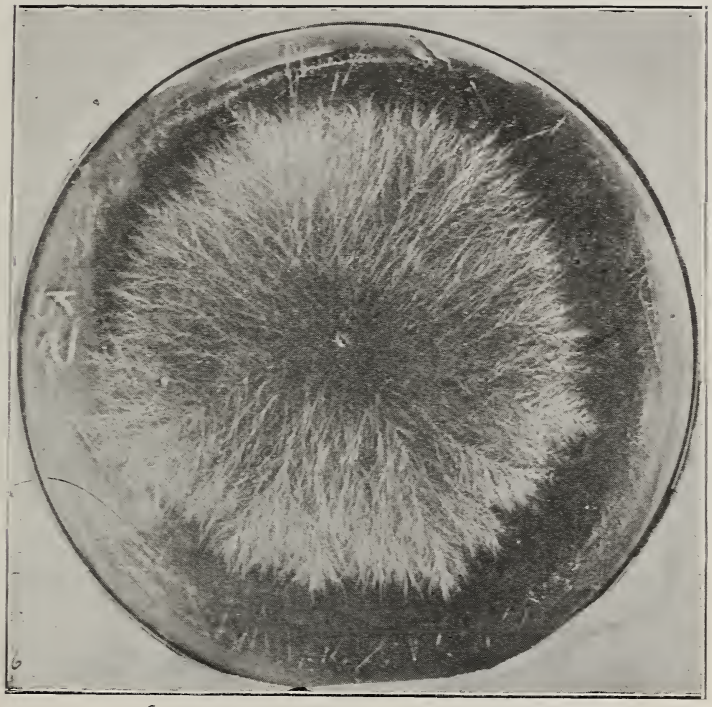

FIG. 16.—Corn-meal agar plate culture of *R. solani*, 2 days old.

FIG. 17.—Mycelium, manner of branching of *R. solani*.

FIG. 19.—*R. palida.* Two-days-old corn-meal agar.

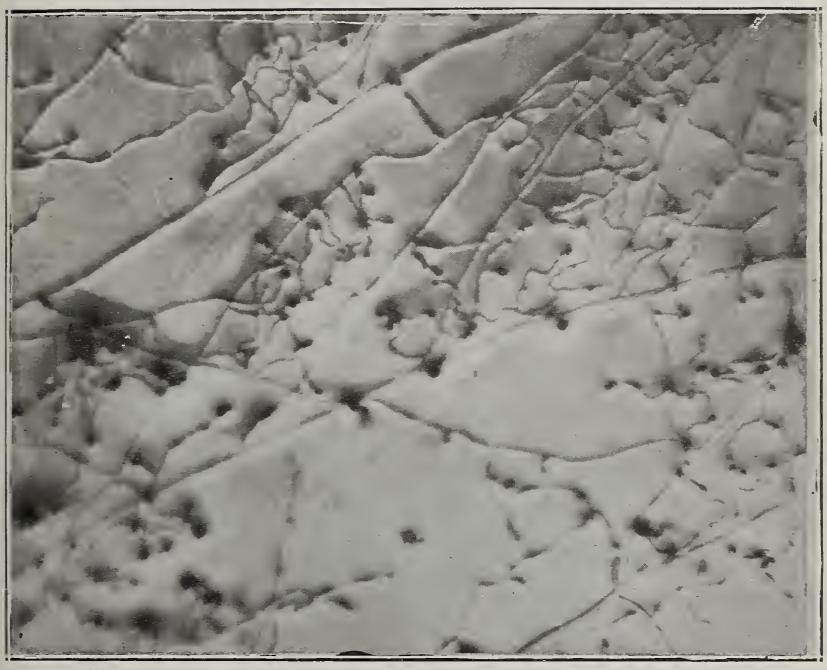

FIG. 20—*R. palida.* **Mycelium. Manner of branching in agar-plate culture.**

FIG. 21.—*R. ferrugina.* Right is a mature culture showing growth characters on sterilized bean pod, and on left a corn-meal agar plate culture three days old.

FIG. 22.—*R. ferrugina.* Mycellum. Manner of branching in corn-meal agar culture.

FIG. 23.—*R. melongena* in the center. On right is a variety of *R. solani* isolated from sugar cane. On left is another *R. solani* from beet. .Note the stringy character of the mycelium in the central tube. All mature cultures.

FIG. 24.—*R. melongena*. Corn-meal ager plate culture two days old.

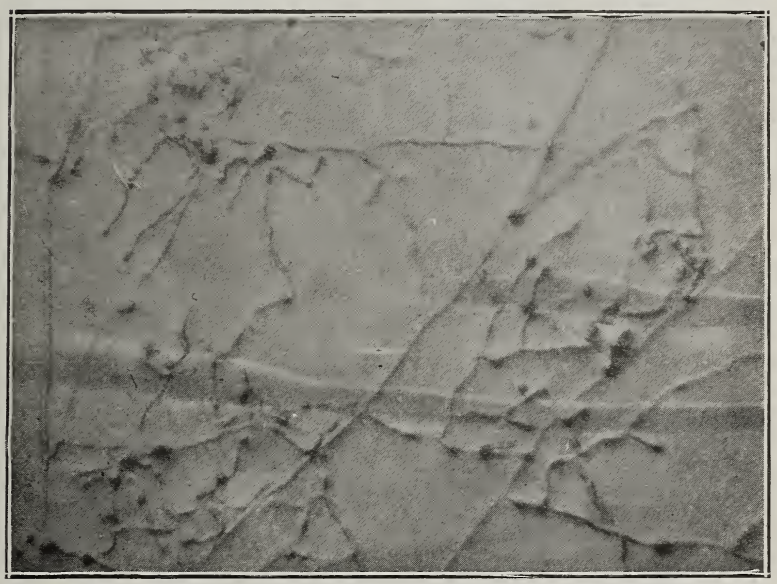

FIG. 25.—*R. melongena.* **Mycellum. Manner of branching in corn-meal agar plate culture.**

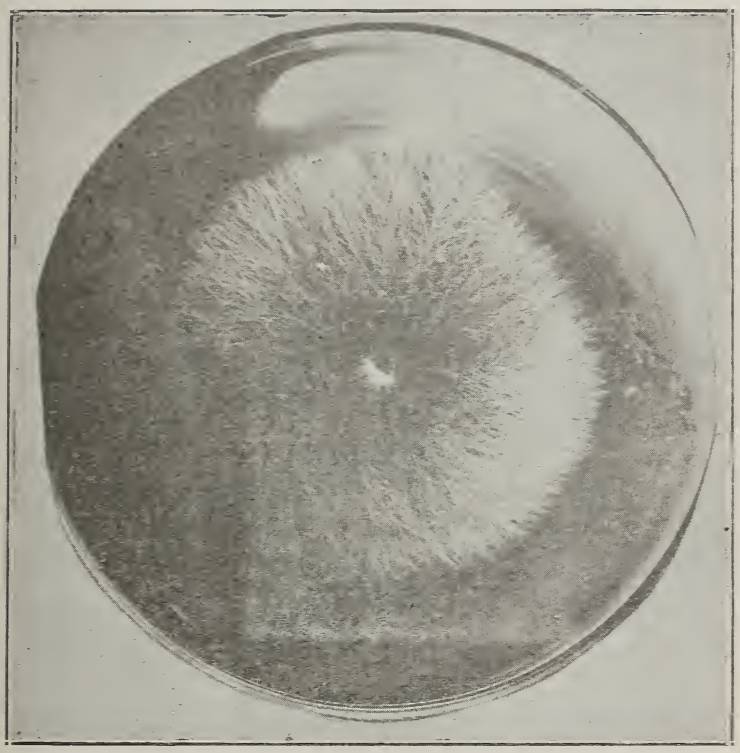

FIG. 27.—*R. alba.* Two-days-old corn-meal agar plate culture.

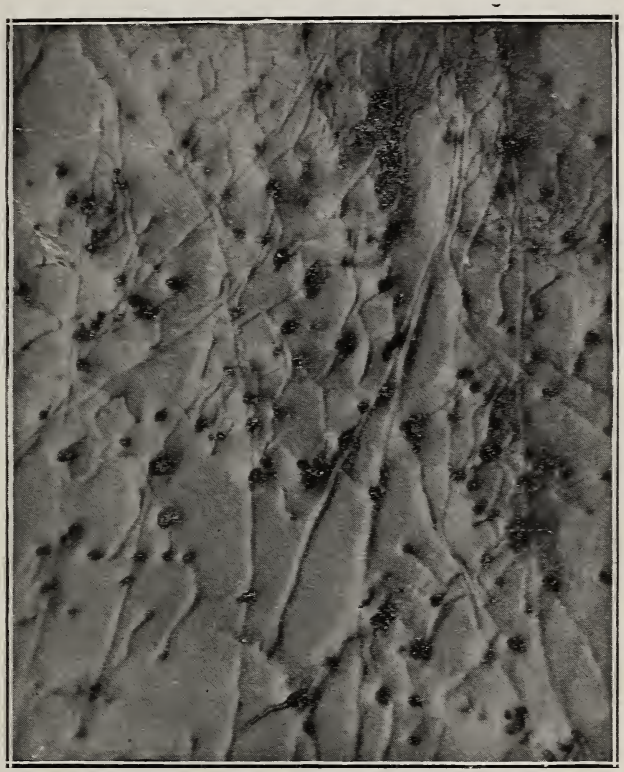

FIG. 28.—*R. alba.* Mycelium. Manner of branching in agar plate culture.